Susceptibility of Eggs and First-Instar Larvae of *Callosamia promethea* and *Antheraea polyphemus* to Malathion[1]

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Abstract. Susceptibility levels to malathion water emulsions are established for *Callosamia promethea* (Drury) and *Antheraea polyphemus* (Cramer): *C. promethea* eggs, LC_{50} = 0.1 mg/ml, probit = 3.3 + 0.9 (log conc); *C. promethea* 1st-instar larvae, LC_{50} = 0.01 mg/ml, probit = 11.3 + 2.9 (log conc); *A. polyphemus* eggs, LC_{50} between 15.6 and 31.2 mg/ml; *A. polyphemus* 1st-instar larvae, LC_{50} = 0.06 mg/ml; probit = 9.9 + 4.1 (log conc); *C. promethea* embryos, LC_{50} = 248 mg/ml, probit = -2.3 + 3.0 (log conc).

Introduction

Giant silkworm moths, such as *Callosamia promethea* (Drury) and *Antheraea polyphemus* (Cramer), used for research purposes, are reared on foliage obtained in agricultural or domestic situations where organophosphate insecticides may be applied or stored. Information is not available on the susceptibility of the early stages of these giant silkworm moths to foliar applications or organophosophate insecticides. To estimate the susceptibility of these insects to natural, potentially contaminated foliage, we determined the laboratory susceptibility of eggs and 1st-instar larvae of *C. promethea* and *A. polyphemus* to malathion, 0,0-dimethyl phosphorodithioate of diethyl mercaptosuccinate, a representative, commonly-used organosphophate material. These estimates of susceptibility are based on laboratory testing procedures, and do not account for field conditions (e.g., pH, temperature, humidity, residue age) that might alter the effective toxicity of the malathion.

Materials and Methods

Test insects were obtained from colonies of *C. promethea* and *A. polyphemus* reared in sleeve cages on wild cherry (*Prunus serotina*) or

[1]Lepidoptera: Saturniidae. This research was not supported by public funds. The opinions contained herein are those of the authors and should not be construed as official or reflecting the views of the Department of the Army.
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various maples (*Acer* spp.) for 10-12 generations. Ortho Malathion 50 Insect Spray[R] (Chevron Chemical Company Ortho Division, San Francisco, CA 94119, EPA Reg. No. 239-739-AC) an emulsifiable concentrate containing 50 percent actual malathion by weight (521 mg/ml), was used for all testing. The appropriate quantity of emulsifiable concentrate was added to water with a volumetric pipet to make a stock emulsion. The stock emulsion was diluted with water, with constant stirring of stock and dilutions, to produce the test concentrations. For larvicide tests, individual hostplant leaves were dipped in test emulsions for 30 seconds and allowed to air dry. Control leaves were dried separately after being dipped only in water. Unfed, 1st-instar larvae (<24 hours old) were placed in random sequence on the treated and control leaves and held in plastic cups and covered with facial tissues for 48 hours before recording mortality. For ovicide tests, egg masses (<48 hours old) were dipped in random sequence in test emulsions for 30 seconds, then allowed to air dry. Control egg masses were dipped in water only, then allowed to air dry separately. Mortality of treated eggs was determined as those that failed to hatch after all control eggs hatched. Embryos of *C. promethea* were tested at high concentrations of malathion (up to 500 mg/ml) to determine the existence of a concentration-mortality relationship. The percentage of embryo mortality rather than the hatch failure was the measured criterion. Mortality data on embryos, eggs, and larvae were subjected to probit analysis using a computer program package described by Barr, et al. (1976).

Results and Discussion

The label recommended concentration for spray application of malathion water emulsion to fruit and ornamental foliage is 12.0 mg AI/ml. Table 1 shows that both eggs and 1st-instar larvae of *C. promethea* and 1st-instar larvae of *A. polyphemus* were highly susceptible to recommended doses. The concentration-mortality relationships for *C. promethea* eggs and 1st-instar larvae are shown in Figures 1 and 2. Eggs of *A. polyphemus* were not susceptible at label concentrations; those exposed to concentrations of up to 15.6 mg/ml showed no ovicidal effects, while those exposed to concentrations of 31.2 mg/ml and above showed no hatch. No concentration-mortality relationship was established for *A. polyphemus* eggs. The concentration-mortality relationship applicable to *A. polyphemus* 1st-instar larvae is shown in Figure 3. Embryos of *C. promethea* were not susceptible to label concentration of malathion water emulsion (Table 1). However, toxicity to embryos was observed at the higher concentrations used in initial range finding. In some of these tests, fully-formed larvae were present in the eggs, but did not hatch. In *C. promethea* the developing dark larva imparts a gray hue to the otherwise white egg. The presence of larvae in the "gray" eggs was confirmed by dissection.

Table 1. Malathion susceptibility of eggs and larvae of *Callosamia promethea* and *Antheraea polyphemus*.[1]

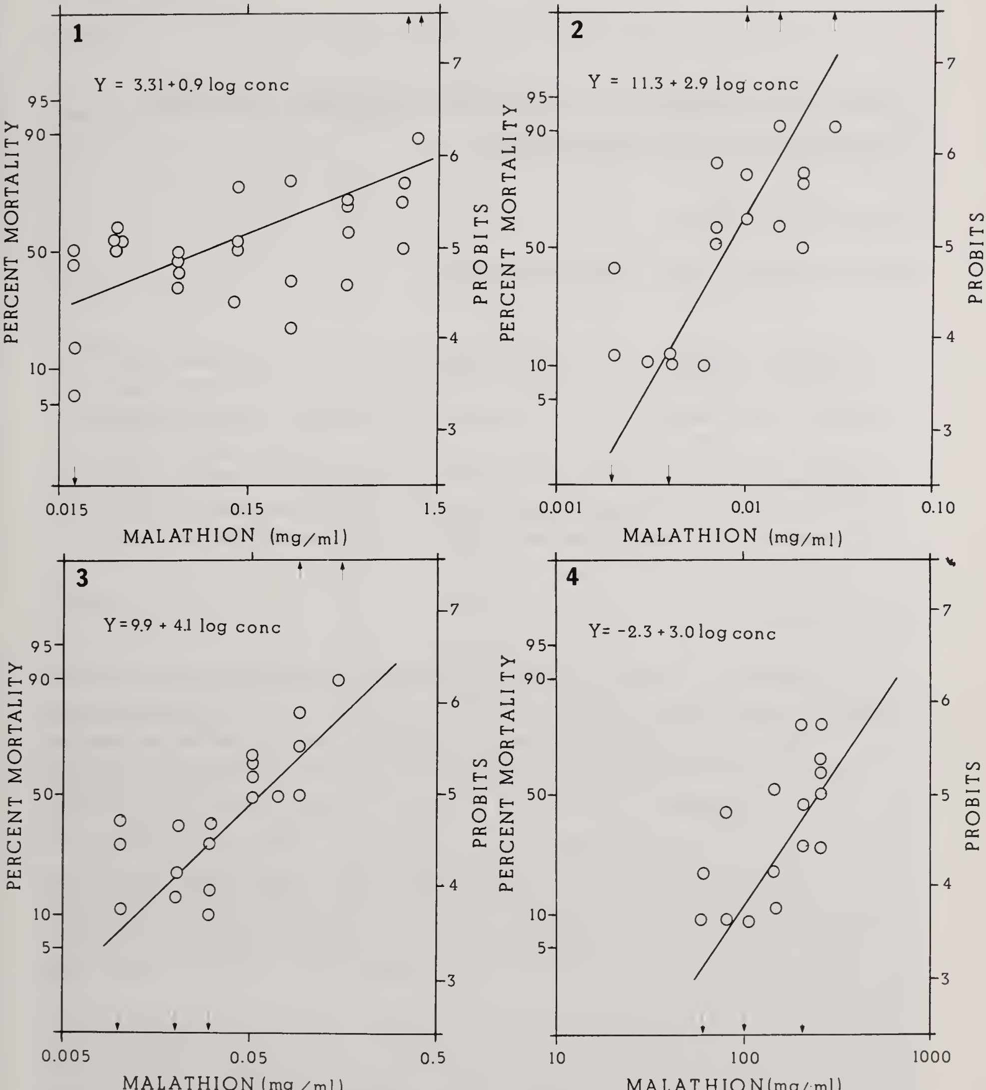
Stage Tested	Number Tested	LC_{50}	LC_{90}	Relative Susceptibility[2] at LC_{50}
		Callosamia promethea		
Embryos	450	248	659	0.05
Eggs	650	0.1	0.36	120
1st-Instar Larvae	380	0.01	0.02	1200
		Antheraea polyphemus		
Eggs[3]	120	>15.6	>15.6	>0.77
1st-Instar Larvae	280	0.06	0.13	200

[1]all values in mg/ml
[2]label concentration/lethal concentration (LC_{50})
[3]no ovicidal effects observed at 15.6 mg/ml

This condition was not observed in ovicide tests with *A. polyphemus* because the light yellow larvae are not visible through the tan egg shells and no dissections were performed. Tests conducted with high concentrations of malathion (Figure 4) demonstrated that a concentration-mortality relationship existed.

Mortality preceding eclosion of eggs has been observed in many lepidopterans in connection with exposure to ovicides, but the phenomenon in general is poorly understood (Smith & Salkeld, 1966). Potter, et al. (1957) reported that high concentrations of TEPP (tetraethyl pyrophosphate) caused mortality in eggs of *Pieris brassicae* Linnaeus; older eggs being considerably more susceptible than younger ones. They concluded that toxicity due to organophosphate exposure appears to involve cholinesterase inhibition at some stage of embryonic development. The *C. promethea* eggs we tested were less than 48 hours old. For those exposed to malathion water emulsion at 0.01 mg/ml all of the embryos developed, but only 50 percent hatched; for those exposed to 248 mg/ml only 50 percent of the embryos developed and none hatched.

These studies establish baseline susceptibility of immature stages of *C. promethea* and *A. polyphemus* to malathion water emulsions, and demonstrate that the immature stages are susceptible at recommended dosages. The particular hazard presented by malathion, or other more toxic or more persistent organosphosphate materials, depends on other variables, such as pH, temperature, humidity, and residue age, not included in these studies.



Figs. 1-4. Susceptibility of Giant Silkworm Moth Eggs and Larvae to Malathion Water Emulsion: 1. *Callosamia promethea* Eggs; 2. *Callosamia promethea* 1st-Instar Larvae; 3. *Antheraea polyphemus* 1st-Instar Larvae; 4. *Callosamia promethea* embryos.

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